```
   1 cgcggccgcgtcgaccgaaaccaggagccgcgggtgttggcgcaaaggttactcccagac
  61 ccttttccggctgacttctgagaaggttgcgcagcagctgtgcccgacagtctagaggcg
 121 cagaagaggaagccatcgcctggccccggctctctggaccttgtctcgctcgggagcgga
 181 aacagcggcagccagagaactgttttaatcatggacaaacaaaactcacagatgaatgct
   1                                 M  D  K  Q  N  S  Q  M  N  A
 241 tctcacccggaaacaaacttgccagttgggtatcctcctcagtatccaccgacagcattc
  11 S  H  P  E  T  N  L  P  V  G  Y  P  P  Q  Y  P  P  T  A  F
 301 caaggacctccaggatatagtggctaccctgggccccaggtcagctacccacccccacca
  31 Q  G  P  P  G  Y  S  G  Y  P  G  P  Q  V  S  Y  P  P  P  P
 361 gccggccattcaggtcctggcccagctggctttcctgtcccaaatcagccagtgtataat
  51 A  G  H  S  G  P  G  P  A  G  F  P  V  P  N  Q  P  V  Y  N
 421 cagccagtatataatcagccagttggagctgcaggggtaccatggatgccagcgccacag
  71 Q  P  V  Y  N  Q  P  V  G  A  A  G  V  P  W  M  P  A  P  Q
 481 cctccattaaactgtccacctggattagaatatttaagtcagatagatcagatactgatt
  91 P  P  L  N  C  P  P  G  L  E  Y  L  S  Q  I  D  Q  I  L  I
 541 catcagcaaattgaacttctggaagttttaacaggttttgaaactaataacaaatatgaa
 111 H  Q  Q  I  F  L  L  E  V  L  T  G  F  E  T  N  N  K  Y  E
 601 attaagaacagctttggacagagggtttactttgcagcggaagatactgattgctgtacc
 131 I  K  N  S  F  G  Q  R  V  Y  F  A  A  E  D  T  D  C  C  T
 661 cgaaattgctgtgggccatctagacctttttaccttgaggattattgataatatgggtcaa
 151 R  N  C  C  G  P  S  R  P  F  T*  L  R  I  I  D  N  M  G  Q
 721 gaagtcataactctggagagaccactaagatgtagcagctgttgttgtccctgctgcctt
 171 E  V  I  T  L  E  R  P  L  R  C  S  S  C  C  C  P  C  C  L
 781 caggagatagaaatccaagctcctcctggtgtaccaataggttatgttattcagacttgg
 191 Q  E  I  E  I  Q  A  P  P  G  V  P  I  G  Y  V  I  Q  T  W
 841 cacccatgtctaccaaagtttacaattcaaaatgagaaaagagaggatgtactaaaaata
 211 H  P  C  L  P  K  F  T  I  Q  N  E  K  R  E  D  V  L  K  I
 901 agtggtccatgtgttgtgtgcagctgttgtggagatgttgattttgagattaaatctctt
 231 S  G  P  C  V  V  C  S  C  C  G  D  V  D  F  E  I  K  S  L
 961 gatgaacagtgtgtggttggcaaaatttccaagcactggactggaattttgagagaggca
 251 D  E  Q  C  V  V  G  K  I  S  K  H  W  T  G  I  L  R  E  A
1021 tttacagacgctgataactttggaatccagttccctttagaccttgatgttaaaatgaaa
 271 F  T  D  A  D  N  F  G  I  Q  F  P  L  D  L  D  V  K  M  K
1081 gctgtaatgattggtgcctgtttcctcattgacttcatgtttttgaaagcactggcagc
 291 A  V  M  I  G  A  C  F  L  I  D  F  M  F  F  E  S  T  G  S
1141 caggaacaaaaatcaggagtgtggtagtggattagtgaaagtctcctcaggaaatctgaa
 311 Q  E  Q  K  S  G  V  W  -
1201 gtctgtatattgattgagactatctaaactcatacctgtatgaattaagctgtaaggcct
1261 gtagctctggttgtatacttttgcttttcaaattatagtttatcttctgtataactgatt
1321 tataaaggtttttgtacatttttttaatactcattgtcaatttgagaaaaaggacatatga
1381 gtttttgcatttattaatgaaacttcctttgaaaaactgctttaaaaaaaagtcgacgcg
1441 gccgc
```

FIG. 1A

```
   1 tctaaagactcaggaaacaaaacctaaattgcctcaaagttcaggtgctttttctccctg
  61 actttagtctagtggagtagtgcagcacctatgcctttctgagaggagtctggagagctg
 121 agtcgctgctggtgctaggattctaggaattcgcctcacttggagctgcatgagaaaaga
 181 aaggcttgcaaatggaggctcctcgctcaggaacatacttgccagctgggtatgccctc
   1                M  E  A  P  R  S  G  T  Y  L  P  A  G  Y  A  P  Q
 241 agtatcctccagcagcagtccaaggacctccagagcatactggacgccccacattccaga
  18   Y  P  P  A  A  V  Q  G  P  P  E  H  T  G  R  P  T  F  Q  T
 301 ctaactaccaagttccccagtctggttatccaggacctcaggctagctacacagtctcaa
  38   N  Y  Q  V  P  Q  S  G  Y  P  G  P  Q  A  S  Y  T  V  S  T
 361 catctggacatgaaggttatgctgctacacggcttcctattcaaaataatcagactatag
  58   S  G  H  E  G  Y  A  A  T  R  L  P  I  Q  N  N  Q  T  I  V
 421 tccttgcaaacactcagtggatgccagcaccaccacctattctgaactgcccacctgggc
  78   L  A  N  T  Q  W  M  P  A  P  P  P  I  L  N  C  P  P  G  L
 481 tagaatacttaaatcagatagatcagcttctgattcatcagcaagttgaacttctagaag
  98   E  Y  L  N  Q  I  D  Q  L  L  I  H  Q  Q  V  E  L  L  E  V
 541 tcttaacaggctttgaaacaaataacaaatttgaaatcaagaacagcctcgggcagatgg
 118   L  T  G  F  E  T  N  N  K  F  E  I  K  N  S  L  G  Q  M  V
 601 tttatgttgcagtggaagatactgactgctgtactcgaaattgctgtgaagcgtctagac
 138   Y  V  A  V  E  D  T  D  C  C  T  R  N  C  C  E  A  S  R  P
 661 ctttcaccttaagaatcctggatcatctgggccaagaagtcatgactctggagcgacctc
 158   F  T  L  R  I  L  D  H  L  G  Q  E  V  M  T  L  E  R  P  L
 721 tgagatgcagtagctgctgcttcccctgctgcctccaggagatagaaatccaggctcctc
 178   R  C  S  S  C  C  F  P  C  C  L  Q  E  I  E  I  Q  A  P  P
 781 cgggggtgccaataggttatgtgactcagacctggcacccatgtctgccaaagctcactc
 198   G  V  P  I  G  Y  V  T  Q  T  W  H  P  C  L  P  K  L  T  L
 841 ttcagaacgacaagagggagaatgttctaaaagtagttggtccatgtgttgcatgcacct
 218   Q  N  D  K  R  E  N  V  L  K  V  V  G  P  C  V  A  C  T  C
 901 gctgttcagatattgactttgagatcaagtctcttgatgaagtgactagaattggtaaga
 238   C  S  D  I  D  F  E  I  K  S  L  D  E  V  T  R  I  G  K  I
 961 tcaccaagcagtggtctggttgtgtgaaagaggccttcacggattcggataactttggga
 258   T  K  Q  W  S  G  C  V  K  E  A  F  T  D  S  D  N  F  G  I
1021 tccaattcccgctagacctggaggtgaagatgaaagctgtgacgcttggtgcttgcttcc
 278   Q  F  P  L  D  L  E  V  K  M  K  A  V  T  L  G  A  C  F  L
1081 tcatagattacatgtttttgaaggctgtgagtaggaacagaaatccgacctgcagtagg
 298   I  D  Y  M  F  P  E  G  C  E  -
1141 aatcaatgaaagaggacagagaagatctgaagtctacacaaggagatcatatgattgaga
1201 gacctggggctttttgatttcttcattgaaatttctcagaatcaagctgttatacatgaa
1261 gcatagtatgtaacattttggttttcaaatggtagtttatcttttacattattggaatag
1321 acctggataattatctttatacacttctaaaaatatgcaccaaattcaagttaaaaaaaa
1381 aaagacgaagagaagtgtatgttttaaaataaaacattttatggaaaagtaagttaaatc
1441 ataatctgggatttatttttcatcttttgttcaatttaaaccttgttagtgctgatttta
1501 ttataaaattgtactttactatcaaacctagttagtttatttcttacagaaatcctccta
1561 ttattttgaaattacatatttttgaaagcttttttaaaagatactattgcctgggaaattc
1621 ta
```

FIG. 1B

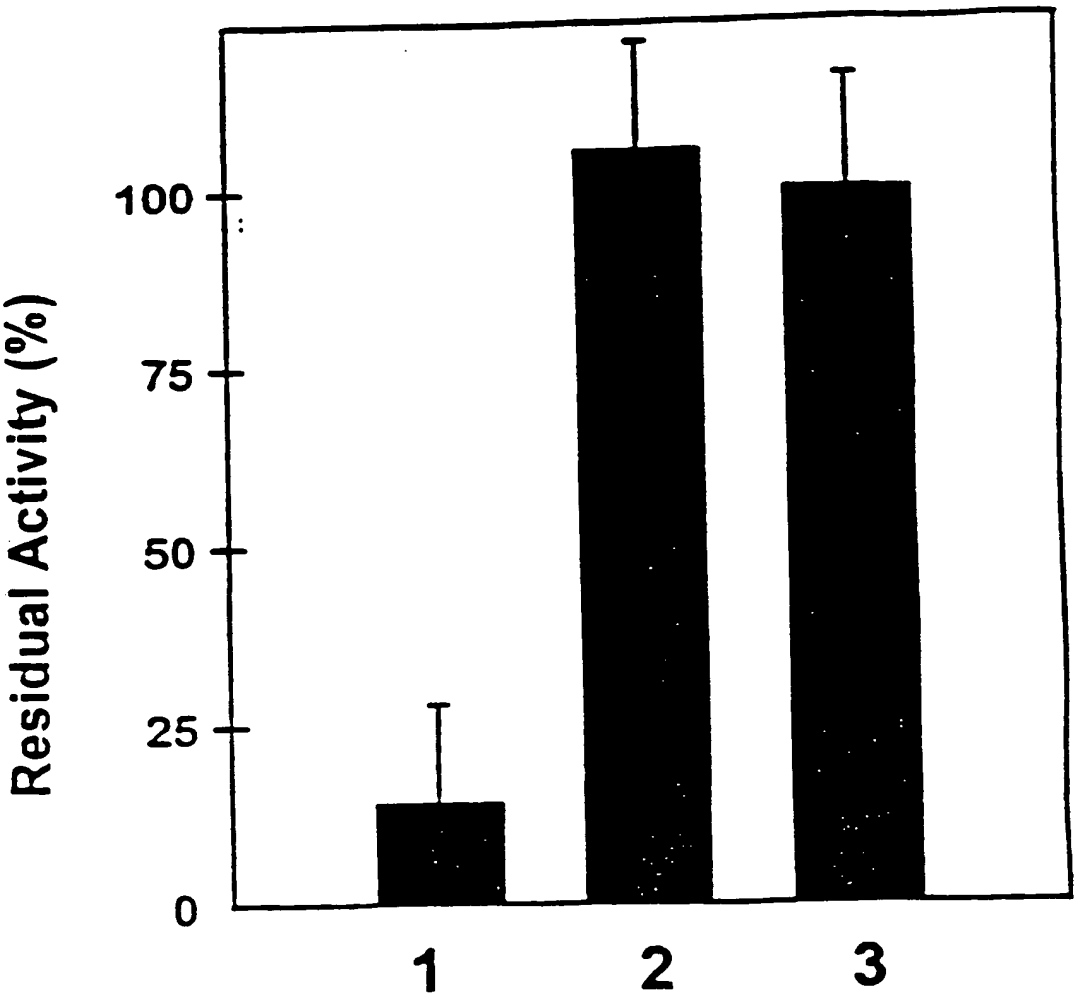

FIG. 2

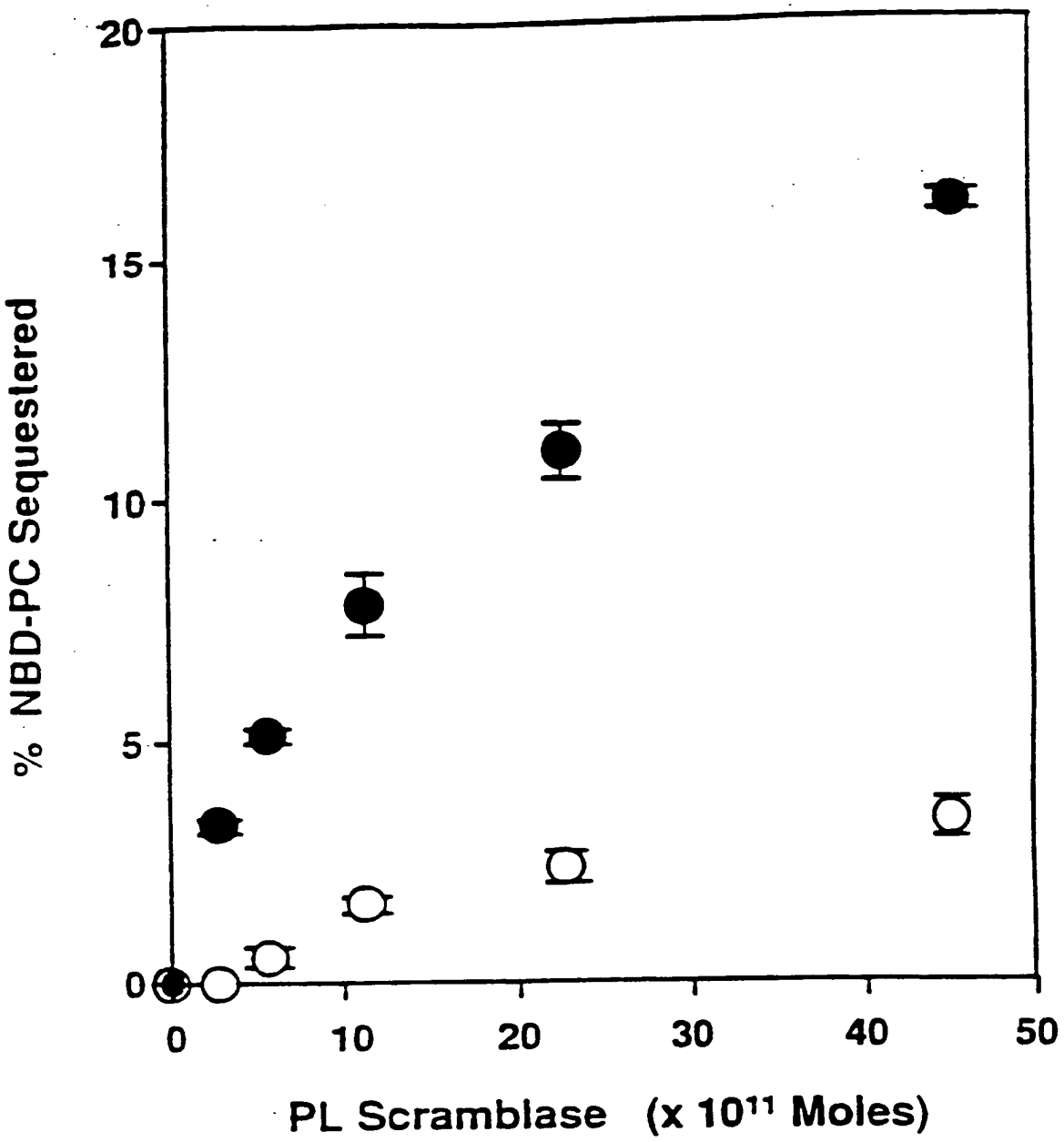
20
15
10
5
0
% NBD-PC Sequestered
0
10
20
30
40
50
PL Scramblase (x 10¹¹ Moles)

FIG. 3

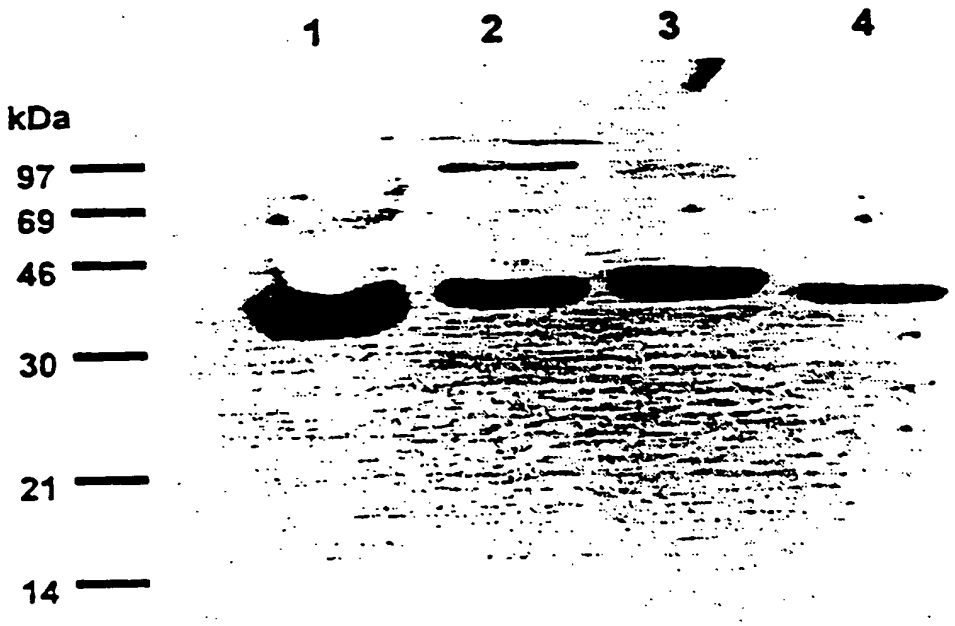

FIG. 4

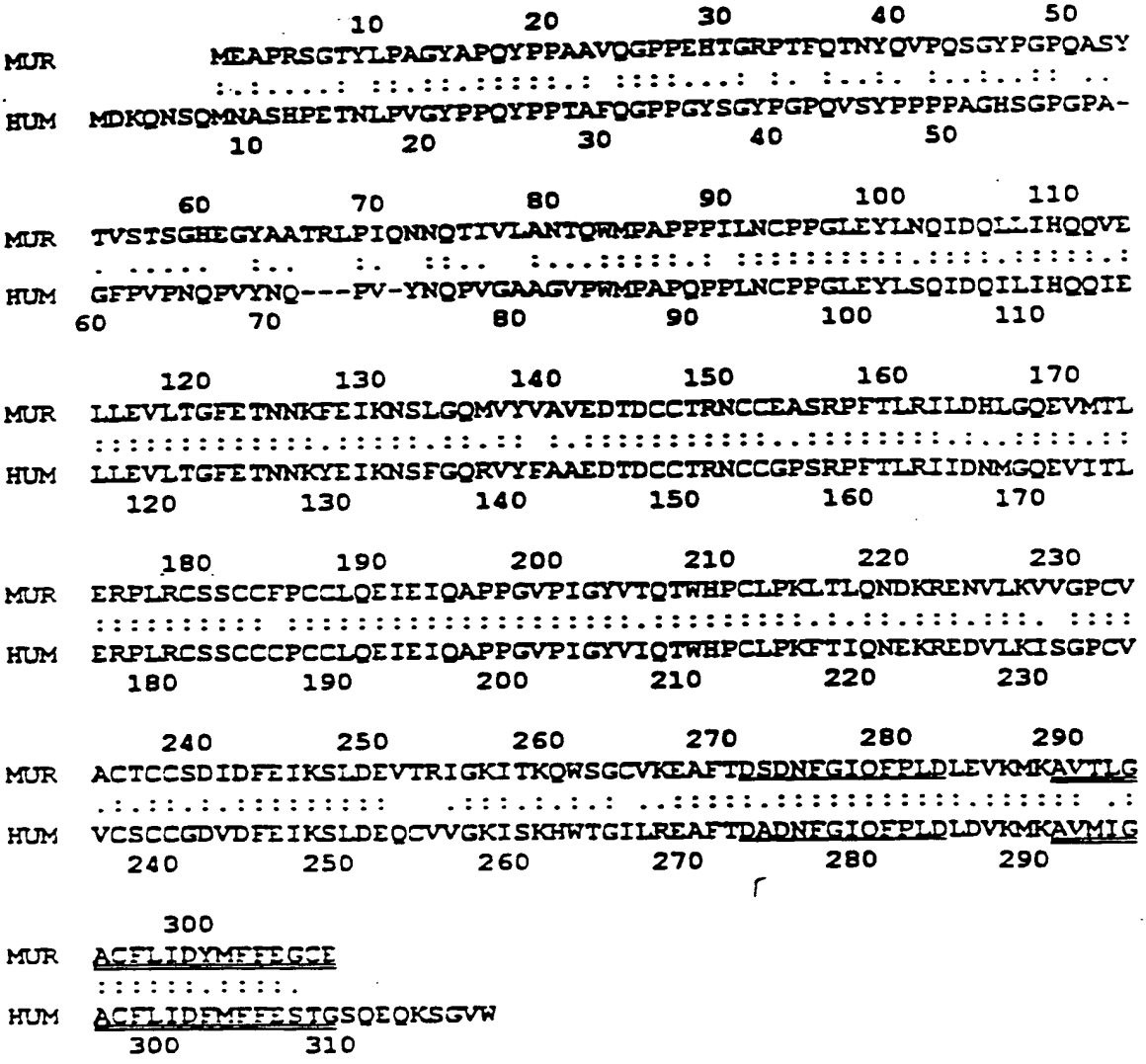
MUR MEAPRSGTYLPAGYAPQYPPAAVQGPPEHTGRPTFQTNYQVPQSGYPGPQASY
HUM MDKQNSQMNASHPETNLPVGYPPQYPPTAFQGPPGYSGYPGPQVSYPPPPAGHSGPGPA-
MUR TVSTSGHEGYAATRLPIQNNQTIVLANTQWMPAPPPILNCPPGLEYLNQIDQLLIHQQVE
HUM GFPVPNQPVYNQ---PV-YNQPVGAAGVPWMPAPQPPLNCPPGLEYLSQIDQILIHQQIE
MUR LLEVLTGFETNNKFEIKNSLGQMVYVAVEDTDCCTRNCCEASRPFTLRILDHLGQEVMTL
HUM LLEVLTGFETNNKYEIKNSFGQRVYFAAEDTDCCTRNCCGPSRPFTLRIIDNMGQEVITL
MUR ERPLRCSSCCFPCCLQEIEIQAPPGVPIGYVTQTWHPCLPKLTLQNDKRENVLKVVGPCV
HUM ERPLRCSSCCCPCCLQEIEIQAPPGVPIGYVIQTWHPCLPKFTIQNEKREDVLKISGPCV
MUR ACTCCSDIDFEIKSLDEVTRIGKITKQWSGCVKEAFTDSDNFGIQFPLDLEVKMKAVTLG
HUM VCSCCGDVDFEIKSLDEQCVVGKISKHWTGILREAFTDADNFGIQFPLDLDVKMKAVMIG
MUR ACFLIDYMFFEGCI
HUM ACFLIDFMFFESIGSQEQKSGVW

FIG. 5

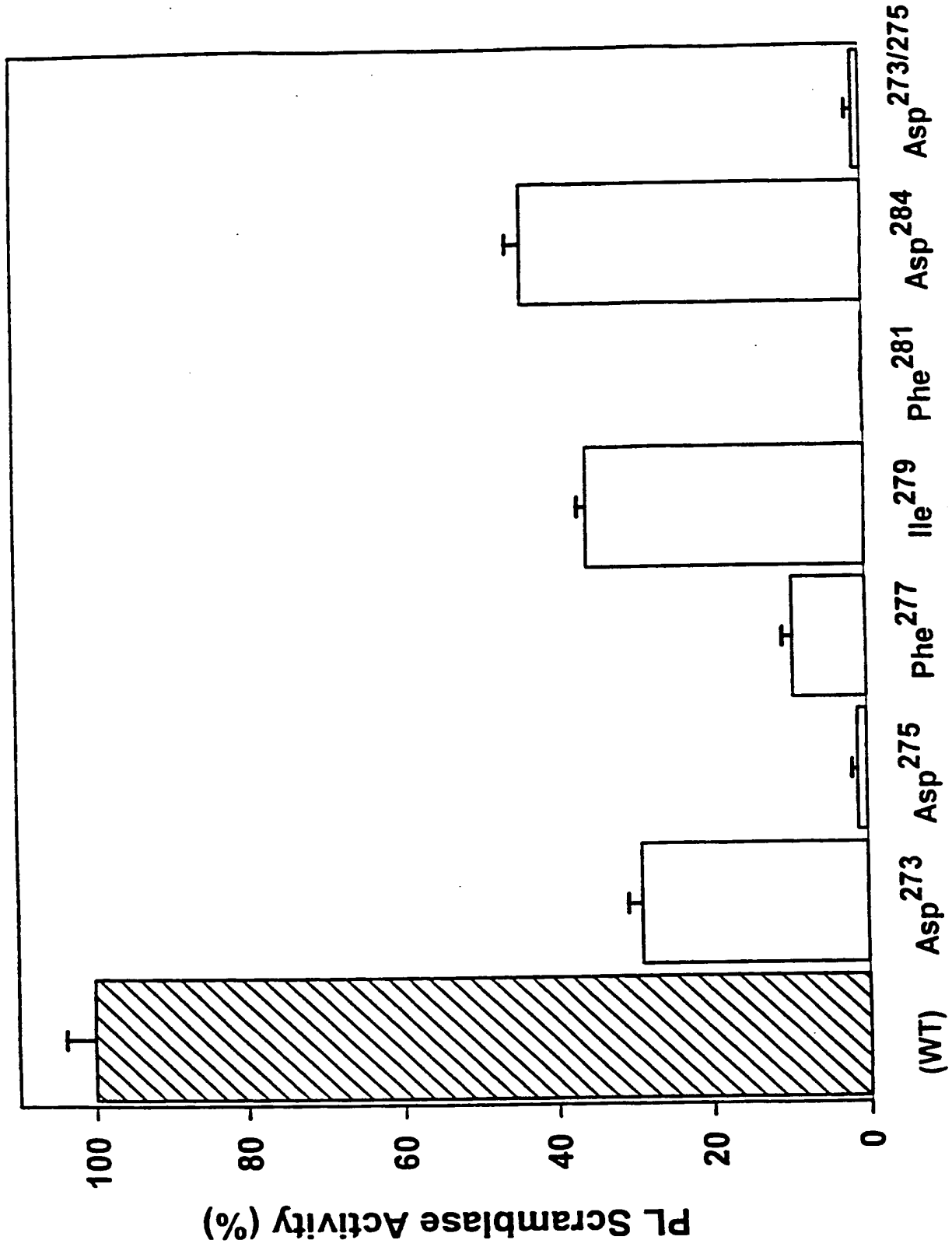
PL Scramblase Activity (%)
0
20
40
60
80
100
(WT)
Asp273
Asp275
Phe277
Ile279
Phe281
Asp284
Asp273/275

FIG. 6

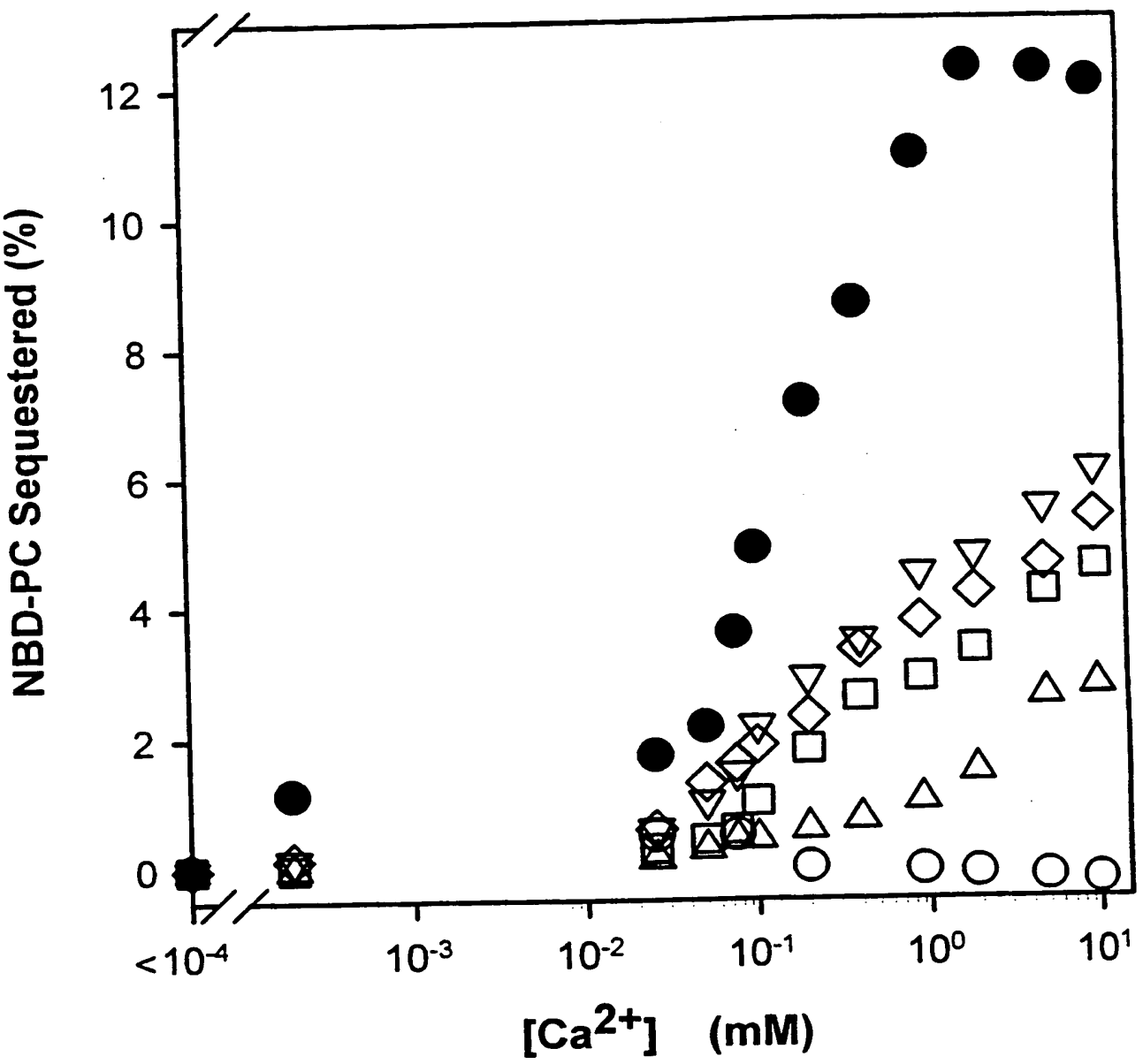

FIG. 7

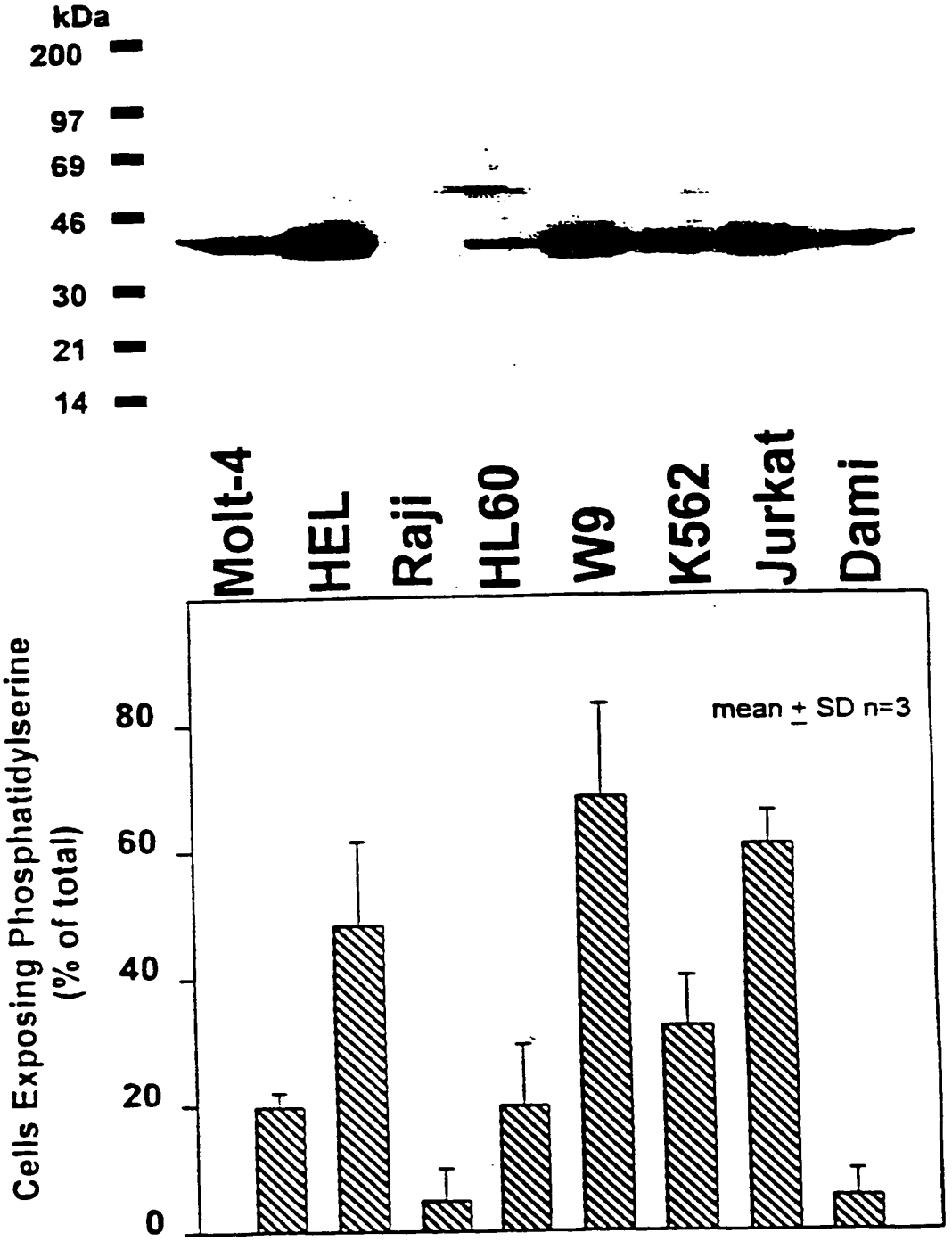
kDa
200
97
69
46
30
21
14
Molt-4
HEL
Raji
HL60
W9
K562
Jurkat
Dami
Cells Exposing Phosphatidylserine
(% of total)
80
60
40
20
0
mean ± SD n=3

FIG. 8

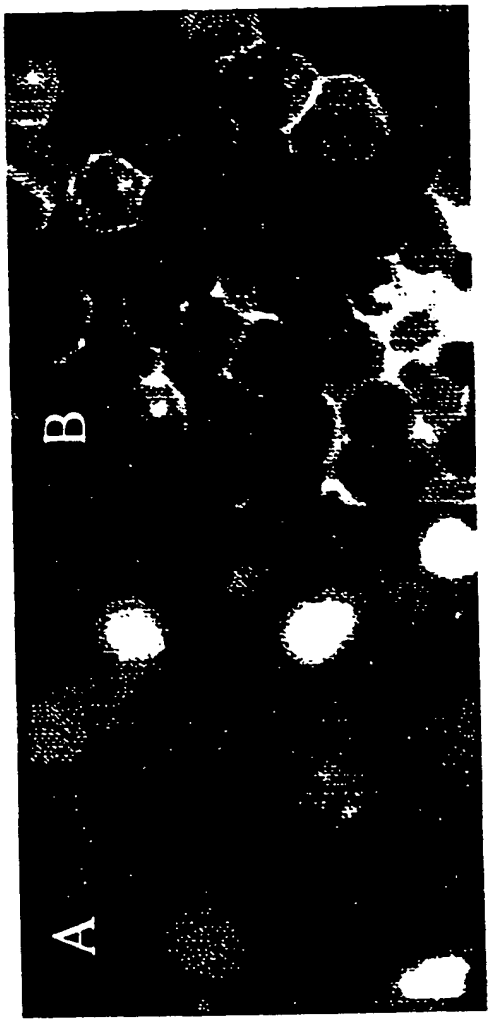

FIG. 9

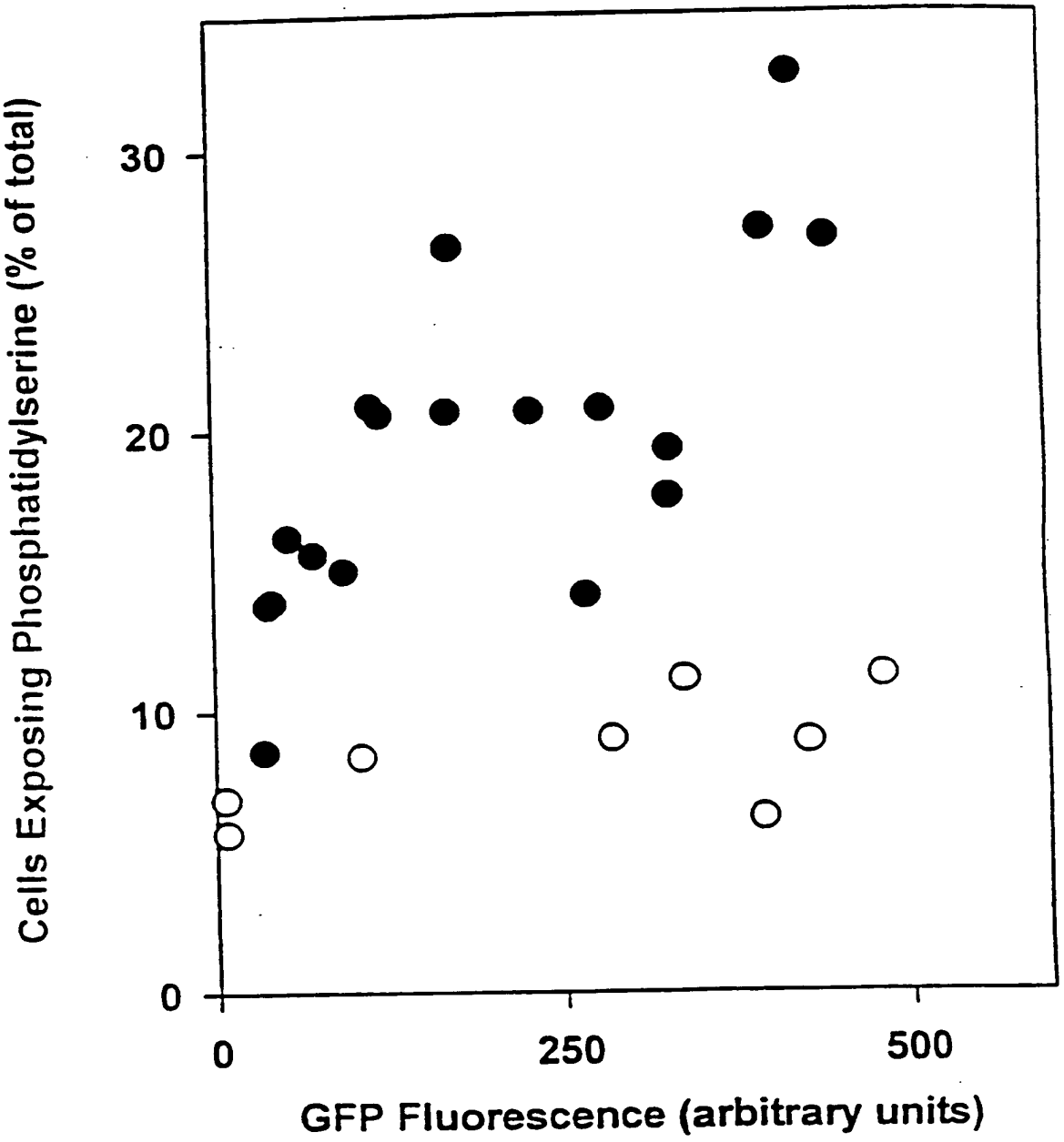
Cells Exposing Phosphatidylserine (% of total)
30
20
10
0
0
250
500
GFP Fluorescence (arbitrary units)

FIG. 10

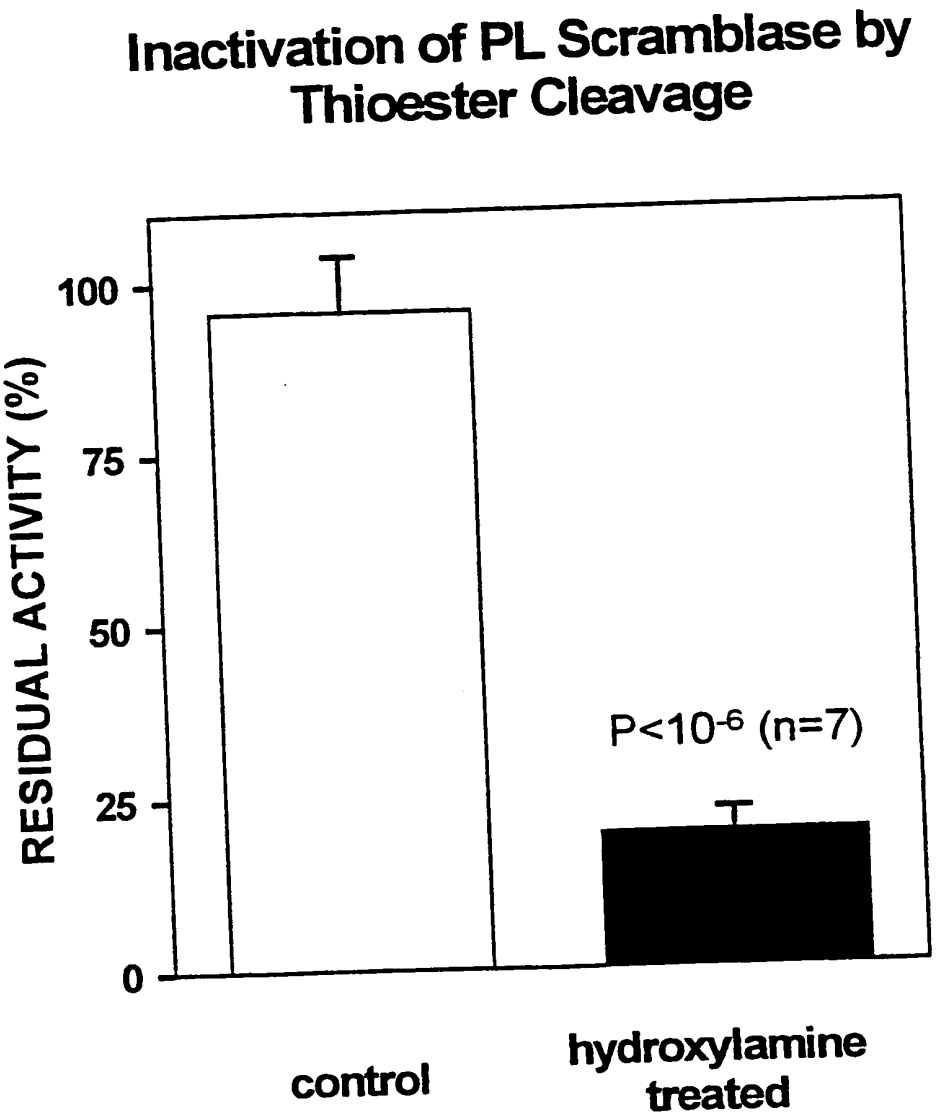
Inactivation of PL Scramblase by Thioester Cleavage
RESIDUAL ACTIVITY (%)
100
75
50
25
0
P<10-6 (n=7)
control
hydroxylamine treated

FIG. 11

Metabolic Labeling of PL Scramblase with [^{3}H]-Palmitate Reveals Covalent Thioester-Linked Fatty Acid

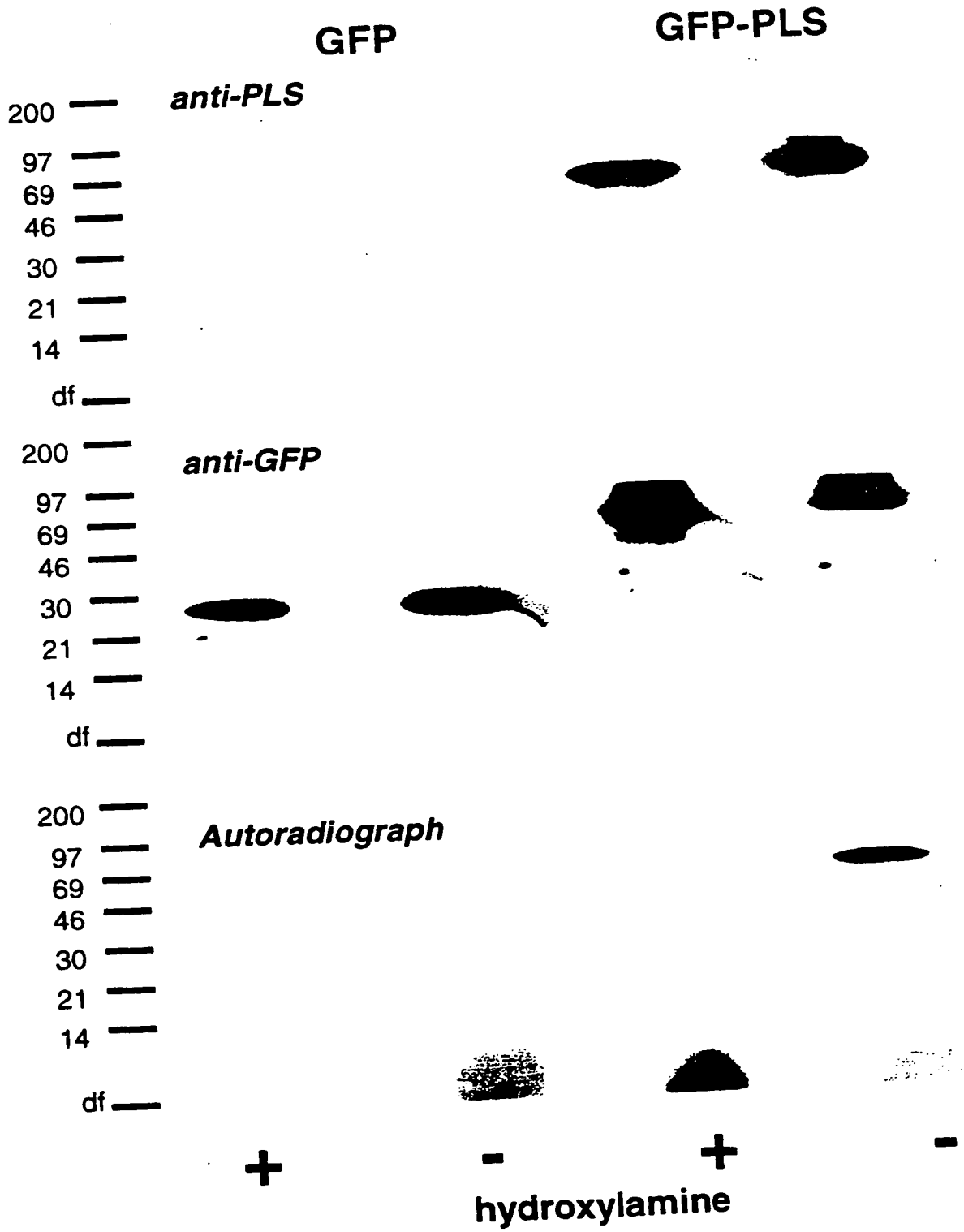

FIG. 12

TLC Analysis of [^{3}H]-Fatty Acid From Hydroxylamine-Treated PL Scramblase

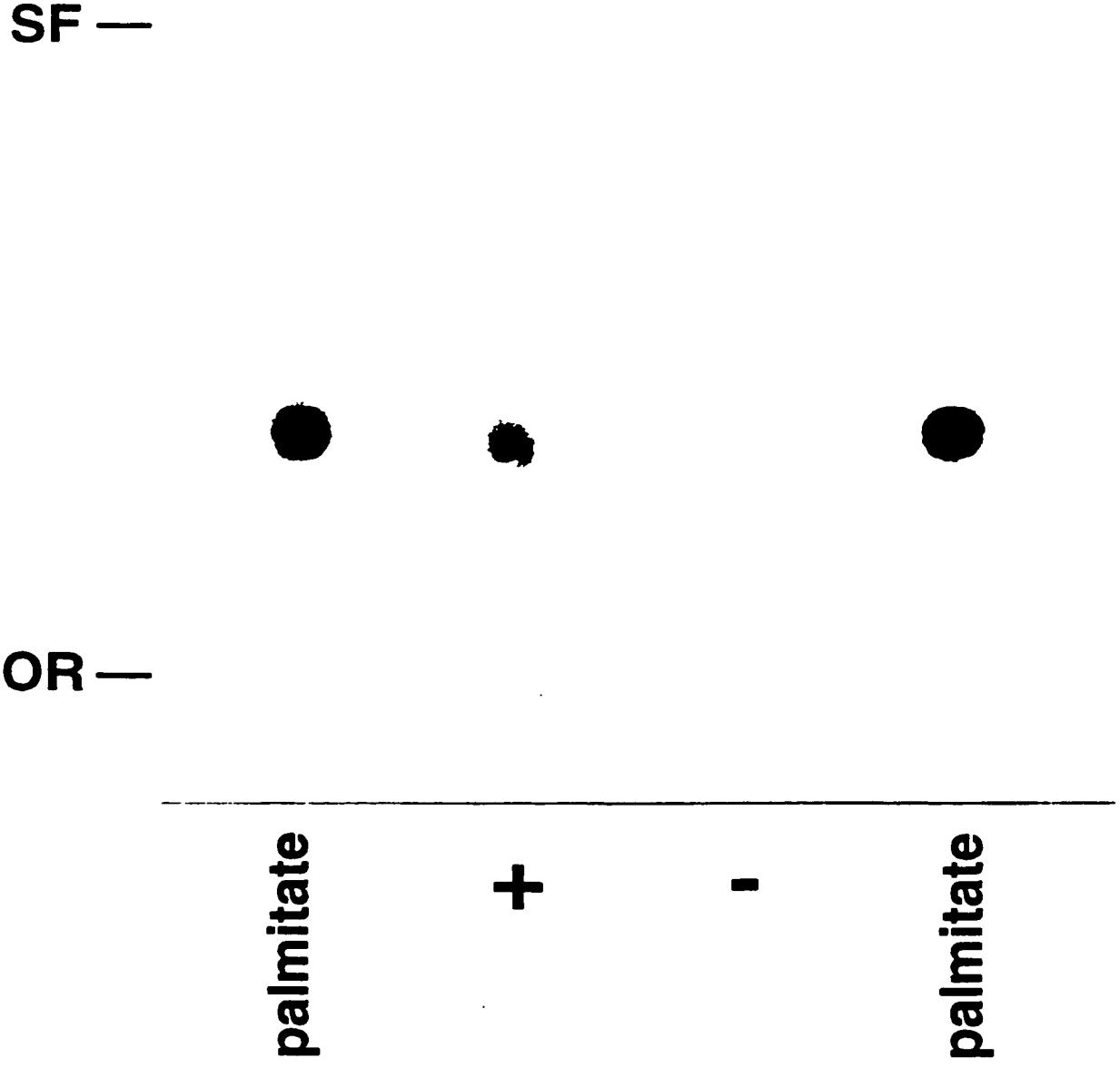

FIG. 13

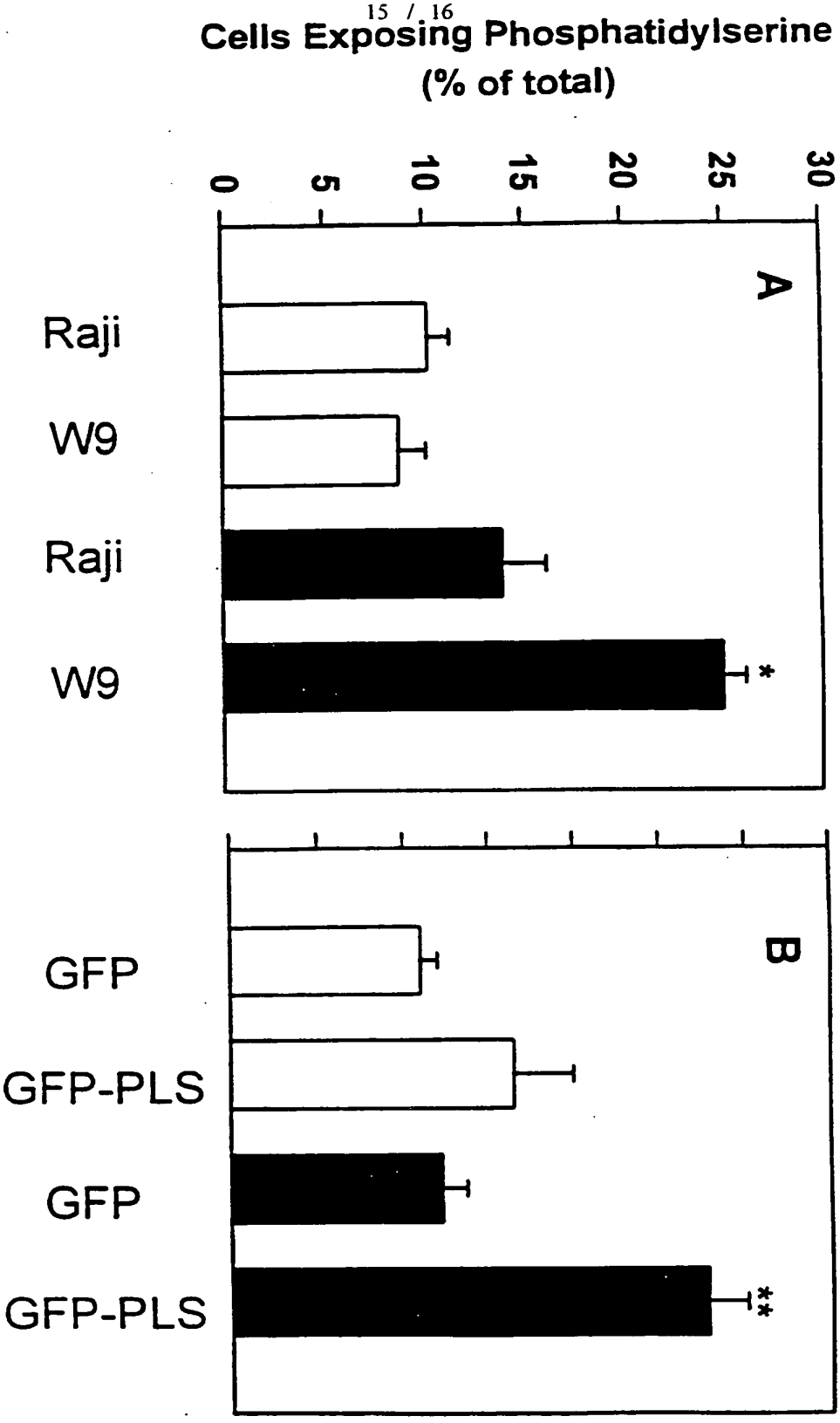
Cells Exposing Phosphatidylserine
(% of total)
0
5
10
15
20
25
30
A
Raji
W9
Raji
W9
*
B
GFP
GFP-PLS
GFP
GFP-PLS
**

FIG. 14

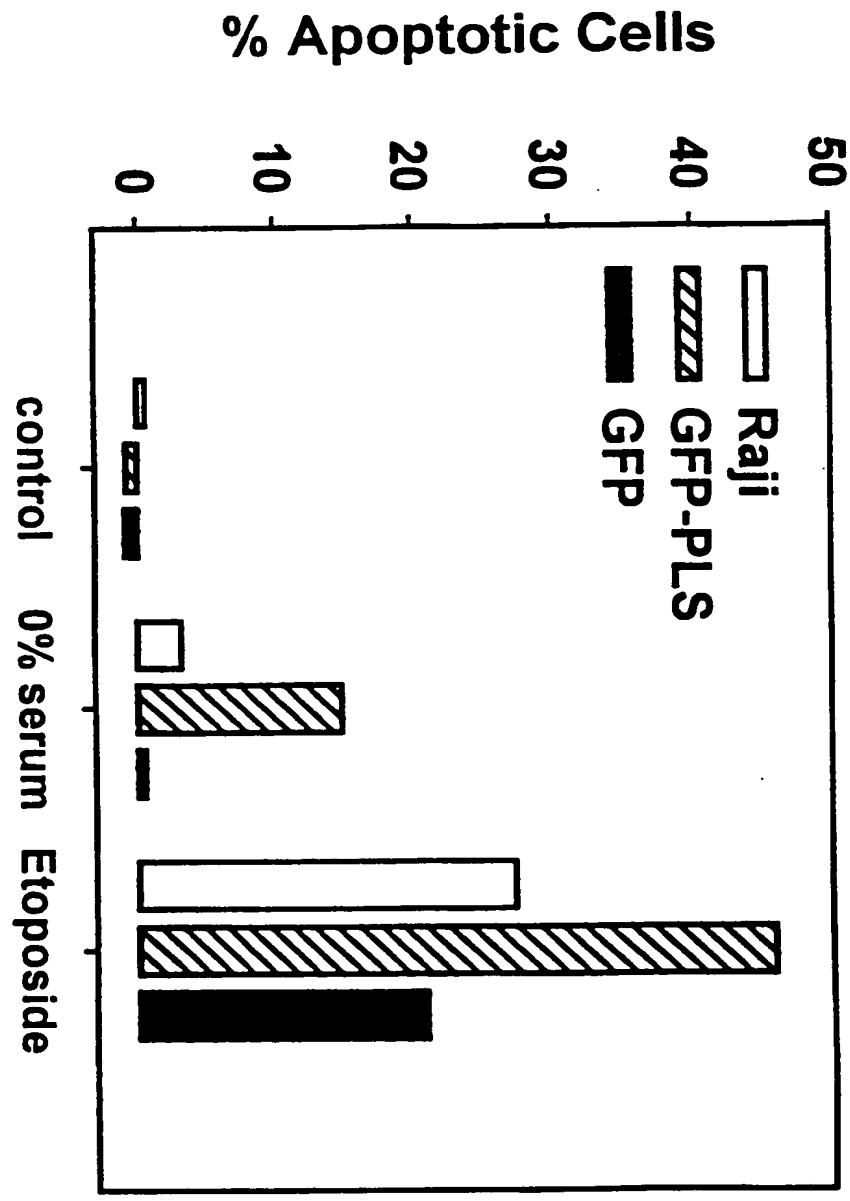
Transfection With cDNA Encoding PL Scramblase
Increases Apoptotic Response of Human B-Lymphoma Line Raji
% Apoptotic Cells
0
10
20
30
40
50
Raji
GFP-PLS
GFP
control
0% serum
Etoposide

FIG. 15